

INVESTIGATION OF PRIMITIVE CHARACTERISTICS OF HUMAN PERIODONTAL LIGAMENT CELLS

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Studies have been demonstrated that human periodontal ligament (PDL) comprises multipotent stem cells. To further delineate the primitive phenotypes of PDL cells, we investigated its morphology, cell surface antigens' expression, and dye efflux activity. After obtaining approval from the Ethical Committee, intact permanent teeth were collected from healthy patients who were undergoing orthodontic treatment at Okayama University Hospital. The PDL cells isolated from these teeth were cultured, then were subjected to following analyses. Primary human PDL cells used in this study showed fibroblastic spindle shape and capillary network formation. Flow cytometric studies demonstrated that PDL cells express mesenchymal stem cell markers, including CD9, CD13, CD29, CD44, CD73, CD90, CD105 and CD166, however, they are negative for many hematopoietic markers (CD2, CD3, CD4, CD8a, CD14, CD16, CD19, CD20, CD24, CD33, CD34, CD38, CD41a, CD45, CD66b, CD117, CD133 and CD235a), endothelial cell markers (Flt-1, Flk-1 and von Willebrand factor), and an epithelial cell marker (cytokeratin 5/8). Integrins positive for PDL cells were CD29 (β 1), CD41a (α 1) and CD51 (α v). Positive cytokine receptors were CD71 (Transferrin R), CD105 (endoglin), CD119 (IFN γ R) and CD120a (TNF RI), and negative were CD117 (c-kit), CD121a (IL-1 R) and CD124 (IL-4 R α). PDL cells expressed matrix receptors such as CD54 (ICAM-1) and CD166 (ALCAM), but lacked CD31 (PECAM-1), CD56 (NCAM) and CD106 (VCAM-1). Human leukocyte antigen (HLA) expressed in PDL cells was HLA-ABC, however, HLA-DR expression was not detected. No reactions of STRO-1 and CDCP-1 (CUB-domain-containing protein 1) were observed in PDL cells. Hoechst dye efflux assay showed that PDL cells include 3.9% SP cells. This SP profile disappeared in the presence of verapamil (50 μ M) or reserpine (5 μ M). Only ABCG2-expressing cells (0.25%) were detected in PDL cells by flow cytometric